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THE ONTOGENY OF SWIMMING BEHAVIOR IN THE SCYPHOZOAN, *AURELIA AURITA*. II. THE EFFECTS OF IONS AND DRUGS[1]

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The active swimming medusa of the scyphozoan, *Aurelia aurita,* develops from a rather inactive, sessile polyp, the scyphistoma. Seasonally, larval medusae (ephyrae) develop by transverse budding of the scyphistoma. This process of medusa formation is termed strobilation, and the scyphistoma at this stage is called a strobila. The ephyrae begin swimming movements while still attached to the strobila. After swimming activity begins, the ephyrae are released to grow and mature into medusae.

Prior to the production of medusae the musculature of a scyphistoma is entirely nonstriated and the nervous system consists of a diffuse nerve net (DNN) functionally arranged in segments (Chapman, 1965, 1966). The development of swimming behavior involves the acquisition of the striated swimming muscles, ganglionic pacemakers, and a through-conducting nerve net which coordinates the swimming beats. The sequence of behavioral and electrophysiological events occurring during the ontogeny of swimming activity are described in the preceding paper (Schwab, 1977), with the conclusion that the coordinating mechanisms involved in swimming are new features of the medusa and not simply modifications of polyp mechanisms. This paper is an investigation undertaken to test this hypothesis by examining the effects of ions and the drugs on the behavior of the scyphistoma, strobila, and adult medusa.

There have been a number of studies examining the effects of ionic variation on the swimming rhythm of scyphomedusae. The effects of the major ions in sea water were determined by using solutions containing those ions in excess of sea water concentration (Mayer, 1906, 1914; Bullock, 1943; Horridge, 1959). The results obtained by these investigators are difficult to interpret since osmotic concentration was not maintained constant. In this study the major ions were reduced or deleted and the osmotic concentration kept constant by substituting another ionic species for the deleted ion.

There have also been many studies on the effects of pharmacological agents on the swimming rhythm of scyphomedusae. Romanes (1877, 1885) observed the effect of various "poisons" (chloroform, "strichnia," curare, and "morphia") on scyphomedusae and found them to have inhibitory effects on the swimming rhythm. Horridge (1959) found that tryptamine accelerates the swimming rhythm of *Cyanea* and *Aurelia* but acetylcholine with and without physostigmine, adrenaline,

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curare, ephedrine, histamine, and 5-hydroxytryptamine had no effect. A substance, as yet unidentified, has been extracted from marginal ganglia of *A. aurita* and found to either increase or decrease spontaneous swimming activity of *A. aurita* depending on the concentration (Barnes and Horridge, 1965). Scyphomedusae, and probably coelenterates as a group, are pharmacologically different from other phyla in being generally unresponsive to common neurohumors (Horridge, 1959; Barnes and Horridge, 1965) and to tetrodotoxin (Mackie, 1968; Schwab, 1972; Ball and Case, 1973).

The approach used in this study is to examine the effects of a number of pharmacological agents and ionic alterations on the behavior of medusae and compare those responses with those of the scyphistoma and strobila. This approach is based on the idea that different coordinating systems in the several life stages might be reflected as different response patterns to ions and drugs. With the response patterns for the medusa established and divided into classes, one or two of the test solutions were selected from each response class and tested on the scyphistoma and strobila. Finding different response patterns would strengthen the hypothesis that coordinating systems of the different life stages are fundamentally different and not simply subtle alterations of the elements present in previous stages.

Materials and Methods

Animals

Medusae of *Aurelia aurita* (5–8 cm in diameter) were collected from the Eel Pond, Woods Hole, Massachusetts. The medusae were kept in a deep tank with slowly flowing sea water until they were used. Polyps of *A. aurita* were obtained from the Supply Department, Marine Biological Laboratory, Woods Hole, Massachusetts. Some additional polyps (Woods Hole strain) were obtained from Dr. Dorothy Spangenberg (University of Colorado, Boulder, Colorado). Scyphistomae were maintained in I⁻-free artificial sea water (ASW) to prevent strobilation (Spangenberg, 1971; for additional details on the maintenance of polyp cultures see Schwab, 1977). Scyphistomae, previously maintained in I-free ASW, strobilate in 6–8 weeks after the polyps are transferred to ASW containing 10^{-5} M KI (Spangenberg, personal communication).

Recording methods

The mechanical activity of swimming medusae was recorded from animals in a constant-temperature chamber containing either sea water (SW) or a test solution maintained at the storage tank temperature (15–18° C). The animals were individually suspended by a hook placed in the aboral mesoglea and connected by silk suture *via* a light spring to a force-displacement transducer. Raising or lowering the transducer adjusted the tension so that the medusa was kept off the bottom of the chamber. The output of each transducer was recorded on a four-channel oscillograph. In order to reduce artifacts resulting from swimming movements, electrical recordings were made from isolated marginal ganglia. Marginal ganglia were removed from medusae and pinned, aboral side up, in a constant temperature chamber adjusted to the temperature of the SW holding tank. A

glass suction electrode with a tip aperture of 50 μm (Schwab, 1977) was attached to the aboral surface of the rhopalium near the ocellus.

Mechanical activity of the tentacles of the scyphistoma was monitored visually or by a photoelectric device and electrical activity was detected with glass suction electrodes (Schwab, 1977). Mechanical activity of the strobila was recorded by observing the tissue and manually deflecting an event marker on a penwriter. Electrical activity from the marginal ganglia of attached ephyrae was recorded by glass suction electrodes.

Electrical potentials from medusae, ephyrae, and scyphistomae were recorded between the glass suction electrode and an Ag/AgCl, indifferent bath electrode. Recorded activity was amplified by a high gain AC-amplifier with a long-time constant and displayed on an oscilloscope and penwriter.

Test solutions

Artificial sea water (ASW) and isosmotic ASW test solutions of various ionic compositions were made from stock solutions prepared with reagent grade chemicals and deionized distilled water (Wilkens, 1970). Cl^--free solutions were prepared with either isethionate or propionate as chloride substitutes. The Cl^--free isethionate solution contained 491.4 mM Na^+, 10.0 mM K^+, 9.8 mM Ca^{+2}, 50.8 mM Mg^{+2}, 65.6 mM SO_4^{-2}, and 491.4 mM isethionate. The concentrations of the ionic species in all other isomotic test solutions are tabulated in Wilkens (1970).

Isosmotic test solutions containing an ionic concentration less than that found in SW were prepared by mixing SW with the ion-free solution in appropriate volumes. The ionic concentration of these test solutions will be expressed as a percentage of the concentration of that ionic species normally found in SW. The test solution containing 108% Na^+ was a 1 : 1 mixture of sea water and 0.54 M NaCl (*i.e.*, 50% Ca^{+2}, 50% K^+, and 50% SO_4^{-2}). The solutions containing pharmacological agents were prepared by dissolving the chemical in SW. The pH and osmotic concentration of all test solutions were determined before use. The solutions were titrated with either NaOH or HCl to pH 7.8 (pH of SW). Osmotic concentrations were measured with a freezing point depression osmometer and adjusted to 925 mOsmol with deionized distilled water.

Experimental design

The protocol used for recording mechanical swimming activity of medusae consisted of a 20 min SW control period, a 20 min test period, and a 20 min SW recovery period. At the end of the control period the SW was removed from the chamber and replaced with 200 ml of the test solution. Following the test period, medusae which ceased to swim were electrically stimulated with platinum pin electrodes, insulated to the tip and inserted into the mesoglea immediately adjacent to the circular swimming muscles. The animals were electrically stimulated once per sec (150 V, 10 msec) for 10 to 15 sec. This gross stimulation determined if the swimming muscles were capable of contracting in the test solution. Following this stimulation, the test solution was drawn off, the chamber and animals washed with 50 ml SW and, finally, the test solution replaced by 200 ml SW.

The mechanical activity in each test solution was analyzed by counting the beats per min (bpm) for each animal during the 20 min control period and the

last 5 min of the test period, determining the difference between them and obtaining the mean difference for the experimental group. The last 5 min of the test period was used in order to obtain the maximum effect of the test solution in the test period. The same control, test, and recovery periods, as well as the solution exchanging procedure used during the mechanical recordings of intact medusae, were also used with isolated ganglia.

Since the tentacles of the polyp are developmentally homologous to the rhopalia of medusae (Thiel, 1966) and are the most electrically active tissue of the polyp (Schwab, 1977), this study was restricted to the differential responses of the tentacles to ions and pharmacological agents.

The same 20 min control period, test period, and recovery period used with medusae was also used with scyphistomae. Electrical and mechanical recordings, however, were done simultaneously rather than sequentially as with medusae. To test the responsiveness of the muscles in the presence of the test solution, the tentacles were electrically stimulated *via* platinum pin electrodes insulated to the tip with teflon. Solutions were added to the test container (see Schwab, 1977) by a gravity fed, polyethylene tube system. As the solution flowed into the test container it overflowed into a larger, outer chamber from whence it was withdrawn by a vacuum line, thus maintaining constant the water level of the test chamber. Fifty ml of a new solution was used to wash out the 1.4 ml volume of the test container. Preliminary experiments had determined that a 10 ml/min rate of flow through the test chamber did not result in any observable response of the polyp to the mechanical stimulus of the changing solution.

The effects of ionic variation and drugs on the electrical and mechanical activity associated with beating activity of ephyrae attached to a strobila were determined by using the same 20 min control, test and recovery period. The methods of recording electrical and mechanical activity, electrical stimulation, and solution exchanging procedure were the same as that used with scyphistomae. All experiments on both scyphistomae and strobilae were done at 4° C in a constant-temperature bath to maintain the animals at the temperature in which they were cultured.

Results

Effects of test solutions on the swimming system of medusae

The effects of test solutions on the mechanical events of swimming are summarized in Table I. Total inhibition of swimming activity was defined as the complete absence of spontaneous swimming activity in all animals tested during the last 5 min in the test solution. Swimming muscles were considered nonfunctional when they did not respond to electrical stimulation during total inhibition of swimming activity. The effects of all solutions tested fell into four types (Table I).

Type I showed no effect on muscles or marginal ganglia. Typical records of the Type I response are shown in Figure 1.

Type II showed total or partial inhibition of swimming beats and marginal ganglion activity, but animals were still responsive to electrical stimulation (Fig. 1). Ten test solutions had a Type II effect on medusae (Table I) and apparently resulted from inhibition of marginal ganglion activity. Intermediate Na^+ concen-

TABLE I

Effects of test solutions on the mechanical activity of the swimming muscles and electrical activity of the marginal ganglia.

Response type	Test solution (isosmotic test solutions from Wilkins, 1970)	Mean difference in beat frequency (beats/min ± s.e.; +, increase; −, decrease in frequency)	N	Total inhibition of spontaneous swimming activity?	Inhibition of MGPs?	Muscle responds to electrical stimulation? (— = not tested)
I	ASW	− 5.3 ± 5.1	12	no	no	—
I	EDTA (2 mM)	+ 2.7 ± 4.0	4	no	no	—
I	EGTA (2 mM)	+ 7.0 ± 4.0	4	no	no	—
I	108% NaCl	−10.8 ± 6.7	4	no	no	—
I	Tyramine (10^{-2} M)	− 3.3 ± 3.6	4	no	no	—
I	Cl^--free (Prop.) ASW	+ 3.0 ± 4.5	6	no	no	—
II	Na^+-free ASW	−31.9 ± 3.6*	6	yes	yes	yes
II	Li ASW	−33.9 ± 2.5*	6	no	yes	—
II	K^+-free ASW	−25.1 ± 1.3*	6	no	yes	—
II	Cl^--free (ISE) ASW	−20.2 ± 3.8*	4	no	yes	—
II	Ca^{+2}-free ASW (+EGTA)	−27.4 ± 3.3*	7	no	yes	—
II	Ca^{+2}-free ASW (NC)**	−38.8 ± 3.4*	8	no	yes	—
II	Ca^{+2}-free Li ASW	−37.6 ± 4.3*	4	no	yes	—
II	Na^+-Ca^{+2}-free ASW (+EGTA)	−27.6 ± 4.2*	4	no	yes	—
II	Na^+-Ca^{+2}-free ASW (NC)**	−36.5 ± 2.6*	4	no	yes	—
II	Na^+-Ca^{+2}-Mg^{+2}-free ASW (+EDTA)	−34.5 ± 5.0*	4	yes	yes	yes
III	Procaine (10^{-2} M)	−33.1 ± 3.2*	6	yes	yes	no
III	Caffeine (10^{-2} M)	−38.2 ± 3.7*	6	yes	yes	no
III	Tryptamine (10^{-2} M)	−30.0 ± 3.8*	4	yes	yes	no
III	Veratrine ($1:10^3$ w/v)	−26.7 ± 0.9*	4	yes	yes	no
IV	Mg^{+2}-free ASW (+EDTA)	+30.5 ± 5.6*	4	no	no	—
IV	Mg^{+2}-free ASW (NC)**	+20.5 ± 4.0*	4	no	no	—
IV	Ca^{+2}-Mg^{+2}-free ASW (+EDTA)	+14.3 ± 2.7*	4	no	no	—
IV	Ca^{+2}-Mg^{+2}-free ASW (NC)**	+18.7 ± 2.8*	4	no	no	—

* $P < 0.05$.
** NC = No chelator.

trations, between 0% Na^+, and 50% Na^+ concentrations, resulted in significant decreases in the rate of swimming activity (Table II).

Type III showed inhibition of swimming beats and MGPs, and the preparation was unresponsive to electrical stimulation (Fig. 1). The effective doses for these compounds are shown in Table III. Characteristically the effect of these drugs, especially at the lower concentrations, was a decrease in contraction amplitude without a change in rate. Similarly, recovery was characterized by an increase in contraction height without a change in rate. In tryptamine (10^{-2} M) or veratrine ($1:10^3$ w/v), medusae failed to recover during the 20 min recovery period; how-

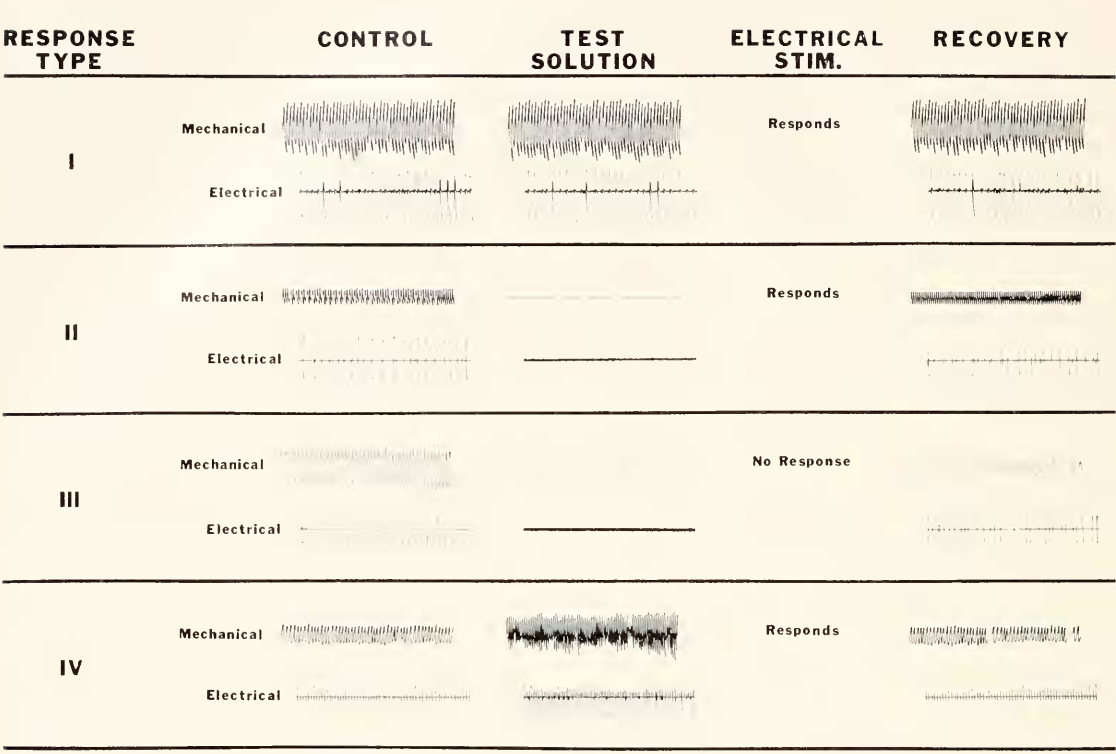

FIGURE 1. Sample records of the mechanical responses of intact medusae (upper traces) and electrical responses of isolated marginal ganglia (lower traces) to test solutions classified according to response types: I, no effect; II, inhibitory effect on pacemaker activity; III, both pacemaker activity and swimming muscles were inhibited; IV, pacemakers excited.

ever, these drugs could be washed out after an extended period in SW. In the case of veratrine, for instance, small contractions began 3 hr after SW replaced the test solution.

TABLE II

Effects of reduced ionic concentrations on the swimming system of A. aurita.

Response type	Test solution (% of SW)	Mean difference in beat frequency (beats/min ± s.e.; +, increase; −, decrease in frequency)	N	Total inhibition of spontaneous swimming activity	Inhibition of MGPs?	Muscle responds to electrical stimulation? (− = not tested)
II	50% Na^+	-39.9 ± 2.5*	4	no	yes	—
II	10% Na^+	-36.7 ± 4.7**	4	yes	yes	yes
II	10% K^+	-26.6 ± 3.6**	4	no	yes	—
II	5% Ca^{+2}	-19.4 ± 4.0*	4	no	yes	—
IV	20% Mg^{+2}	$+22.4 \pm 3.5$*	4	no	no	—
IV	10% Mg_+^2	$+22.9 \pm 4.2$*	3	no	no	—

* $P < 0.05$.
** $P < 0.01$.

TABLE III

Effects of drugs on the swimming system of A. aurita.

Response type	Test solution	Mean difference in beat frequency (beats/min ± s.e.; +, increase; −, decrease in frequency)	N	Total inhibition of spontaneous swimming activity?	Inhibition of MGPs?	Muscle responds to electrical stimulation? (− = not tested)
III	10^{-2} M Procaine	−33.1 ± 3.2*	6	yes	yes	yes
III	5×10^{-3} M Procaine	−31.7 ± 2.7*	6	yes	yes	yes
II	10^{-3} M Procaine	−24.2 ± 3.9*	6	no	yes	—
II	5×10^{-4} M Procaine	− 9.1 ± 3.2*	4	no	yes	—
III	10^{-2} M Caffeine	−38.2 ± 3.7*	6	yes	yes	yes
III	5×10^{-3} M Caffeine	−32.1 ± 2.5*	6	yes	yes	yes
II	2.5×10^{-3} M Caffeine	−19.7 ± 3.1*	6	no	yes	—
II	10^{-3} M Caffeine	− 7.3 ± 2.4*	6	no	yes	—
III	10^{-2} M Tryptamine	−25.9 ± 3.8**	4	yes	yes	yes
II	10^{-3} M Tryptamine	−15.2 ± 1.9**	4	no	yes	—
III	$1:10^3$ Veratrine	−26.7 ± 0.9***	4	yes	yes	yes
II	$1:10^4$ Veratrine	−27.4 ± 4.9***	4	no	yes	—

* $P < 0.05$.
** $P < 0.01$.
*** $P < 0.001$.

Type IV showed an increased rate of mechanical activity. This was seen in solutions lacking Mg^{+2} or with reduced Mg^{+2} concentrations (Table II). In SW the appearance of the bell alternates between a disk (relaxed) and a bell (swimming contraction). During high frequencies of contraction, as in Mg^{+2}-free ASW, activity was erratic and the animal assumed a constant bell shape due to incomplete relaxations (Fig. 1, Type IV). Sometimes this response was more pronounced and the medusa lost coordination; many contractile events could be seen occurring independently in different sectors of the bell. The mechanical activity returned to normal following replacement of the test solution with SW (Fig. 1, Type IV). Similarly, isolated ganglia showed a high rate of activity in both Mg^{+2}-free and Ca^{+2}-free ASW (Fig. 1, Type IV). The increased swimming activity in Ca^{+2}-Mg^{+2}-free ASW is surprising, since reducing Ca^{+2} alone is inhibitory. Apparently the absence of Ca^{+2} cannot reverse the excitatory effect in the absence of Mg^{+2}.

One possible response pattern, inhibition of swimming activity without affecting MGP activity (*i.e.*, uncoupling muscle contractions from GFNN activity), was not seen with any of the test solutions.

Effect of test solutions on the tentacular system of polyps

The tentacular system responded quite differently than the swimming system to the solutions tested (Table IV). Procaine, which inhibits both pacemaker activity and the swimming muscles of medusae and Mg^{+2}-free ASW, which leads to hyperexcitability in medusae, had no effect on spontaneous electrical activity and tentacle movements in polyps. In medusae, Na^{+}-free ASW inhibits marginal pacemaker output in the swimming system but in the polyp both TCPs and tentacle contractility are inhibited. The swimming system and the tentacular system responded similarly

TABLE IV

Differential responses of the three stages of the life cycle of Aurelia aurita *to test solutions.*

	Response type			
	I (No response)	II (Inhibition of pacemaker activity)	III (Inhibition of pacemakers and swimming muscles)	IV (Increase in pacemaker output)
Swimming system of the medusa	ASW	Na^+-free ASW Ca^{+2}-free Li ASW	Procaine (10^{-2} M)	Mg^{+2}-free ASW
Tentacular system of the polyp	ASW Procaine (10^{-2} M) Mg^{+2}-free ASW	Ca^{+2}-free Li ASW	Na^+-free ASW	
Beating system of the strobila	ASW Procaine (10^{-2} M) Mg^{+2}-free Li ASW		Na^+-free ASW	Mg^{+2}-free ASW

to Ca^{+2}-free ASW. Typical responses of the tentacular system to the test solutions are shown in Figure 2.

Effects of test solutions on the beating system of attached ephyrae

The effects of the test solutions on the beating system are shown in Figure 3. ASW, procaine (10^{-2} M), and Ca^{+2}-free Li ASW had no effect on the beating

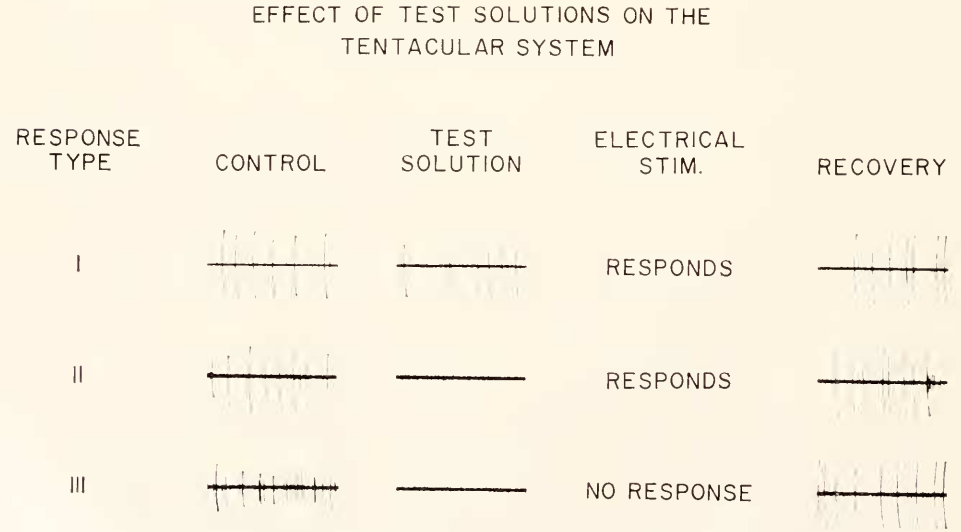

FIGURE 2. Effect of test solutions on the tentacular system classified according to the response types established for the medusa: I, no effect; II, only the TCPs were inhibited; III, both TCPs and the muscles were inhibited.

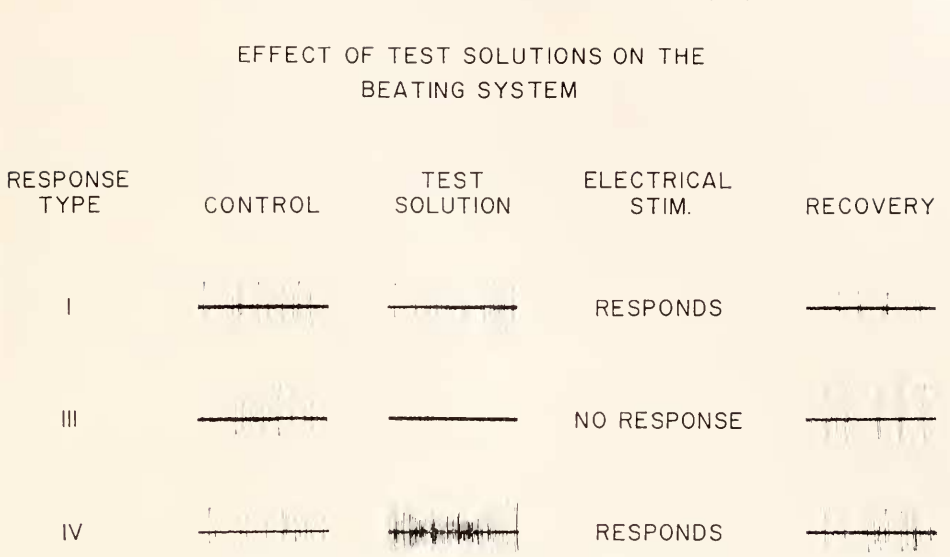

FIGURE 3. Effect of test solutions on the beating system classified according to the response types established for the medusa: I, no effect; III, both pacemakers and beating muscles were inhibited; IV, pacemakers excited.

system. Na^{+}-free ASW inhibited both electrical and beating activity while Mg^{+2}-free ASW caused an increase in the rate of beating (Table IV).

DISCUSSION

Most of the test solutions had either no effect (Type I) or an inhibitory effect on swimming activity of medusae which resulted from an inhibition of pacemaker activity (Type II) or an inhibition of both pacemakers and swimming muscles (Type III). Four test solutions increased swimming activity (Type IV response). This increased rate of swimming in the absence of Mg^{+2} was surprising, although adding excess Mg^{+2} to sea water is a commonly used anaesthetic for marine invertebrates. *Haliclystus auricula,* a sessile scyphozoan, does not exhibit the swimming contractions normally associated with medusae. Stauromedusae, such as *H. auricula* have been considered to be derived from scyphistomae which failed to completely develop into free medusae (Hyman, 1940). Therefore, residual pacemaker activity has been suspected, but none has been demonstrated (Gwilliam, 1960). Residual pacemaker activity might be detected in the absence of Mg^{+2}; however, *H. auricula* failed to show any excitatory effects thus providing additional evidence for the complete lack of pacemaker activity in these animals. In addition, the hydromedusae, *Sarsia tubulosa* and *Aequorea aequorea,* do not respond with an increase in swimming activity in the absence of Mg^{+2} (Schwab, unpublished observations).

The increased rate of swimming activity in Mg^{+2}-free ASW was caused by an increase in the rate of pacemaker firing. The absolute refractory period of the swimming muscles in sea water is approximately 0.7 sec (Bullock, 1943; Pantin and Vianna Dias, 1952), which corresponds to a maximum possible rate of

85 bpm. The highest stimulation frequency to which a ganglion-free muscle preparation will respond in a one-to-one fashion is 1.3 pulses/sec (Bullock, 1943) or 78 bpm. The rate of activity caused by the test solutions did not exceed the upper limits determined from the control animals (approx 80 bpm). Therefore, the increase in rate probably resulted from pacemaker excitation only, and a decrease in the refractory period of the muscles need not be postulated.

Many investigators have subjected coelenterates to media containing various pharmacologically active compounds. For instance, the sympathomimetics, tryptamine and tyramine, were found to initiate contractions in the anemone, *Calliactis*, but adrenaline was without effect (Ross, 1945, 1957). In the hydroid, *Corymorpha*, tyramine increases electrical activity (Ball and Case, 1973). The medusae of *A. aurita* responded differently to both of these compounds. Tyramine was totally without effect on the swimming system of *A. aurita*, yet tryptamine inhibited both pacemakers and swimming muscles (*contra* Horridge, 1959). Veratrine, a mixture of alkaloids, causes contracture in vertebrate skeletal muscle and spontaneous contractions in *Calliactis* (Ross, 1945) but, at the same concentration, did not cause contracture of the swimming muscles; rather it inhibited both pacemakers and swimming muscles. Caffeine, which also causes contracture in vertebrate skeletal muscle, stimulates vertebrate cardiac muscle and excites the central nervous system, had none of these effects on the medusa of *A. aurita* but rather caused the same inhibition as tryptamine, veratrine, and procaine. Tetrodotoxin (TTX) and procaine both block the action potential in squid axons (Nakamura, Nakajima, and Grundfest, 1965; Taylor, 1959). TTX had no effect on the swimming system (Schwab, 1972) but procaine, previously thought to affect only the swimming muscles (Schwab, 1972), blocks both the swimming muscles and the MGPs. Similarly, TTX has also failed to block electrical activity in the hydroids *Corymorpha* (Ball and Case, 1973) and *Cordylophora* (Mackie, 1968). Many other compounds, effective in other systems (*e.g.*, acetylcholine, adrenaline, histamine, curare, ephedrine, and 5-hydroxytryptamine) are also without effect on the scyphozoan swimming system (Horridge, 1959). This evidence shows that the coelenterate neuromuscular system is pharmacologically quite different from those in other phyla, and further, that the scyphozoan neuromuscular system is pharmacologically quite different from other classes of Cnidaria.

There are several differences between the effects of ionic variation on the swimming activity of *A. aurita* and those reported earlier for other species of medusae (Mayer, 1906; Horridge, 1956a, b). For instance, excess Ca^{+2} totally inhibited swimming activity in *Cassiopea andromeda* and *C. xamanchana* but only partially inhibited swimming activity in *A. aurita*. Excess Na^{+} totally inhibited swimming activity of *C. xamanchana*, whereas excess Na^{+} had no significant effect on the activity of *A. aurita*. Since no information is available on the ionic mechanisms underlying cnidarian nervous activity, these differences, at the moment, cannot be explained. The results also cannot eliminate the possibility that the test solutions which inhibited or stimulated pacemaker output may have affected the inputs to pacemakers rather than pacemakers directly. The exact site of each effect and the mechanism behind the effects remains unknown.

Regardless of mechanism, the responses obtained from the medusa of *A. aurita* are useful as an index of physiological maturity when compared with the responses obtained from other stages of the life cycle. For instance, Mg^{+2}-free ASW, which

greatly increased swimming activity in medusae and beating activity in ephyrae, had no effect on the polyp. The excitatory effect of Mg^{+2}-free ASW on the ephyra supports the hypothesis that the excitatory effect of Mg^{+2}-free ASW is general to the pacemakers found in swimming scyphozoan forms. Thus the development of the Type IV response may be considered as the development of a medusoid response. The Type III response to procaine was specific to the adult medusa and is also considered a medusoid response. Physiological development is not complete in the ephyra, since the Type III response to procaine was not observed. Although the comparison of the responses between the polyp, strobila, and medusa show no other clear relationships, the limited number of test solutions used was sufficient to establish that the tentacular system, beating system, and swimming system are physiologically different.

In summary, the transformation of the sessile polyp to an active swimming medusa involves the development of both active pacemakers interconnected by a fast, through-conducting nerve net and striated swimming muscles. Both develop during strobilation causing the ephyra to exhibit swimming behavior similar to the medusa. Perhaps this similarity between the medusa and ephyra previously obscured the similarities between the polyp and ephyra. Now there is evidence that the newly acquired medusoid nerve net and accompanying behavior in the strobila (*i.e.*, ephyra) is superimposed over a polypoid nerve net and behavior (Schwab, 1977). The evidence presented here suggests that the strobila is not only a developmental mixture of both polypoid and medusoid behavioral characteristics but also physiological characteristics. In spite of obvious similarities, the beating system of the ephyra and the swimming system of the medusa are not physiologically or behaviorally identical. Therefore, as the ephyra matures into an adult, morphological maturation must, perforce, be accompanied by further physiological maturation of the neuromuscular system responsible for producing the swimming movements.

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Summary

1. The responses of *Aurelia* medusae to pharmacological agents and ionic variation were classified into four response types: Type I, no response; Type II, inhibition of pacemaker activity; Type III, inhibition of both pacemakers and swimming muscles; and Type IV, increase in pacemaker output.

2. The swimming pacemakers of *Aurelia* medusae become hyperactive in Mg^{+2}-free solutions (Type IV). This response appears to be general in swimming scyphozoa.

3. The response pattern to pharmacologically-active compounds indicates that the coelenterate neuromuscular system is quite different than those in other phyla. In fact, the response spectrum is not consistent within the Cnidaria.

4. Similarly, the responses of adult medusae to ionic variation show no consistent pattern within various scyphomedusae.

5. Test solutions from each response type established with medusae were selected and tested on the scyphistoma and strobila stages. The comparison of

the responses to the test solutions between the medusa, scyphistoma, and strobila showed that the neuromuscular systems are physiologically different. The strobila, specifically the ephyra, is a mixture of both polypoid and medusoid response types. The strobila, therefore, is physiologically an intermediate stage in the development of the adult medusa.

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